NORMAL SKIN REACTIONS TO ULTRAVIOLET LIGHT

1. An Attempt to Modify Normal Erythema and Pigmentation with Methoxsalen*

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Much has been written about the psoralens and ultraviolet light (1). It has been suggested by several investigators that suntanning may be augmented through ingestion of 8-methoxypsoralen (2, 3) and that other responses of normal human skin to solar radiation can be altered by ingestion of these compounds (4).

The following experiments were devised to determine the effect of ultraviolet light on the skin of normal human volunteers taking methoxsalen (8-methoxypsoralen). They were conducted during the winter months using artificial sources and again in the summer on clear sunny days between 10:00 A.M. and 2:00 P.M., when the subjects were exposed to natural sunlight.

The sources of artificial radiation were as follows:

(1) A Westinghouse fluorescent sun-lamp, emitting a continuous spectrum of rays, 2900–3500 A. U. (designated as the FS40T-12 lamp).

(2) A standard hot-quartz ultraviolet lamp emitting a line spectrum with an intense band at 2967 A. U.

(3) A Carbon-arc lamp ("C" carbon) emitting a continuous spectrum.

In order to explore these effects more thoroughly, various schedules of drug administration were used in combination with the ultraviolet light exposures from these different sources.

The ultraviolet light exposures also were varied in their intensity and frequency of administration. Finally, various time relationships between drug ingestion and ultraviolet light exposure were employed.

Erythema resulting from ultraviolet light exposures within the sunburn spectrum is graded usually as 1+, 2+, 3+ and 4+.

1+ Erythema is barely perceptible erythema. The amount of exposure required to produce a 1+ erythema 24 hours after exposure is designated as the minimal erythema dose (M.E.D.)

4+ erythema is the vivid maximal degree erythema produced by an ultraviolet light exposure. The least amount of exposure required to produce a 4+ erythema, 24 hours after exposure, is designated as the "minimal 4+ erythema dose."

Increased exposure beyond the minimal 4+ erythema dose does not alter the degree of visible erythema, but may result in increased inflammatory reaction manifested by edema and vesiculation.

Intermediary grades, 2+ and 3+ erythema, were estimated.

All erythema readings were made independently by two observers.

EXPERIMENT #1

Purpose

To determine the effect of a *single* dose of methoxsalen on the minimal erythema dose of ultraviolet light from artificial sources.

Method

Thirty (30) normal healthy white males received graded exposures of the rays from a carbon arc lamp ("C" carbon) and a Westinghouse fluorescent sun-lamp (FS40T12) to test sites on the back, to determine the minimal erythema dose (MED) for each subject. These readings were graded visually.

Twenty (20) of these individuals were given a single dose of methoxsalen. Five men received 5 mg.; 5 received 10 mg.; 5 received 20 mg. and 5 received 40 mg. The other ten (10) subjects received placebo medication.

The minimal erythema dose was determined again for each subject on different test sites 2 hours after drug ingestion and again 24 hours later.

Results

A single dose of methoxsalen did not change the amount of ultraviolet light required to pro-

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duce minimal erythema in any subject in this group.

EXPERIMENT #2

Purpose

To determine the effect of *repeated doses* of methoxsalen on the minimal erythema dose of ultraviolet light, from artificial sources.

Method

Thirty (30) normal healthy white males received graded exposures of the rays from a carbon arc lamp ("C" carbon) and a fluorescent sun-lamp (FS40T12) to test sites on the back. The MED for each individual was determined as in experiment 1.

Twenty (20) subjects received methoxsalen daily for one month. Five received 5 mg. daily; 5 received 10 mg.; 5 received 20 mg. and 5 received 40 mg. The MED was determined each week on different skin sites for the four weeks that the drug was administered, and also each week for the next four weeks.

Ten (10) subjects served as controls and were given placebo tablets. They received the same ultraviolet light exposures as received by subjects taking methoxsalen.

Results

Repeated doses of methoxsalen did not alter the amount of ultraviolet light required to produce minimal erythema in any subject in this group.

EXPERIMENT #3

Purpose

To determine the effect on erythema and pigmentation of methoxsalen administered daily with subsequent *daily* ultraviolet light exposures from artificial sources.

Method

The MED was determined for 30 normal healthy white males using a carbon arc lamp and a fluorescent sun-lamp. Twenty (20) of these individuals received methoxsalen daily for one week in the following amounts: 5 received 5 mg. daily; 5 received 10 mg.; 5 received 20 mg. and 5 received 40 mg. The remaining 10 subjects were given placebo medication. Each day, two hours after drug or placebo ingestion, a minimal erythema exposure was given to the *same* 5 cm. square area of the upper back.

TABLE I EXPERIMENT #3

Daily dosage of methoxsalen

Degree of Pigmentation	5 mg.	10 mg.	20 mg.	40 mg.	Placebo
0	1				
+1	2		2		2
+2		2	3	2	3
+3	1	3		2	3
+4	1			1	2

Degree of pigmentation resulting from repeated exposures of ultraviolet light to the same skin site. (Figures indicate the number of subjects receiving drug dose indicated).

At the end of the week, the minimal erythema dose also was determined on a skin site *NOT* previously exposed to the artificial light sources during the course of this experiment.

Results

(1) Repeated daily minimal erythema exposures of ultraviolet light to the same skin site did not produce significant differences in the erythema reaction in subjects taking methoxsalen as compared with the control group.

(2) Repeated daily minimal erythema exposures of ultraviolet light to the same skin site produced intense pigmentation in some individuals and lesser degrees of pigmentation in others. These gradations of pigmentation occurred whether the subject received placebo, or methoxsalen, and appeared to be unaffected by methoxsalen ingestion. (table I).

(3) There was no alteration in the minimal erythema dose in a previously unexposed site either in subjects receiving methoxsalen or in control subjects.

(4) There was no correlation between the degree of pigmentation in the site not previously exposed in individuals receiving varying doses of methoxsalen, or placebo.

EXPERIMENT #4

Purpose

To determine the effect of *repeated doses* of methoxsalen on the minimal erythema dose of ultraviolet light, from natural sunlight.

This experiment duplicated Experiment #2,

with the exception that summer sunlight during June and July was used as the source of ultraviolet light.

Results

No change was observed in the minimal erythema dose. in any of the subjects in this group.

EXPERIMENT #5

Purpose

To determine the effects on erythema and pigmentation, of methoxsalen administered daily, with subsequent daily minimal erythema exposures of light to the same site from natural sunlight. This was a repetition of Experiment #3, using natural summer sunlight instead of artificial light sources.

Results

The findings were comparable to those in Experiment #3.

EXPERIMENT #6

Purpose

To determine the effect of daily sunlight exposures for 7 days during July, in subjects taking methoxsalen *daily*.

Method

Twenty (20) subjects received methoxsalen each day in the following amounts: 5 received 5 mg.; 5 received 10 mg.; 5 received 20 mg. and 5 received 40 mg. Ten control subjects received placebo.

All were given 45 minute daily mid-day sunlight exposures to the anterior abdominal wall.

Results

There was no significant correlation between the amount of erythema and pigmentation observed in subjects receiving various amounts of methoxsalen or placebo, when they were exposed to natural sunlight daily for one week, (table II).

TABLE II

Results after one week of daily exposure to sunlight in patients taking medication for one week

Subject	Daily Dose of Methoxsalen	Erythema	Pigmentation
1	5 mg.	3+	3+
2	5 mg.	1+	3+
3	5 mg.	1+	3+
4	5 mg.	2+	2+
5	5 mg.	1+	1+
6	10 mg.	—	2+
7	10 mg.	4+	1+
8	10 mg.	4+	2+
9	10 mg.	2+	1+
10	10 mg.	1+	2+
11	20 mg.	—	1+
12	20 mg.	4+	1+
13	20 mg.	3+	2+
14	20 mg.	1+	1+
15	20 mg.	2+	3+
16	40 mg.	4+	—
17	40 mg.	4+	1+
18	40 mg.	3+	2+
19	40 mg.	1+	3+
20	40 mg.	—	3+
21	Placebo	3+	1+
22	Placebo	1+	1+
23	Placebo	—	1+
24	Placebo	4+	1+
25	Placebo	1+	2+
26	Placebo	1+	3+
27	Placebo	—	1+
28	Placebo	2+	3+
29	Placebo	4+	1+
30	Placebo	1+	2+

EXPERIMENT #7

Purpose

To determine the effect of a *single* dose of methoxsalen on erythema and pigmentation when reexposed to sunlight, 1 month after initial exposure.

Twenty (20) men received a single dose of methoxsalen (5 received 5 mg.; 5 received 10 mg.; 5 received 20 mg.; 5 received 40 mg.). Ten (10) men served as controls and received a single dose of placebo. Two and half hours later, one-half of the abdomen of each subject was exposed to a 4+ erythema dose of sunlight. Further exposures to sunlight were not permitted.

One month later the pigmentation on this previously exposed area was recorded. Then, the *entire* abdomen of each subject was exposed to a 4+ erythema dose of sunlight. The results were observed the following day and again one month later.

Results

(1) Although each subject developed an initial 4+ erythema on the test side, one month later different amounts of residual pigmentation were noted in various subjects ranging from 0 to 4+ both in the placebo group *and* the drug treated group. However, there was no correlation between the degree of pigmentation remaining, and the initial dosage of methoxsalen or the placebo.

(2) Although reexposure of the entire abdomen of each subject resulted in a 4+ erythema on the previously unexposed side, the degrees of erythema produced on the previously *exposed* side varied from 0 to +4, with no correlation between the erythema produced in the placebo treated group as compared to the drug treated group. There was also no correlation between the degree of erythema and dose of methoxsalen.

EXPERIMENT #8

Purpose

To determine the effect of daily 4+ erythema doses of sunlight in patients taking methoxsalen daily for one week.

Method

Twenty (20) men received methoxsalen daily for one week (5 received 5 mg.; 5 received 10 mg.; 5 received 20 mg.; 5 received 40 mg.). Ten (10) men served as controls and received a daily dose of placebo.

All subjects received a 4+ erythema exposure from sunlight 2½ hours after ingestion of the medication, to one half of the abdomen each day during the week they were taking the medication. One month later, the pigmentation on this previously exposed area was noted. Then the entire abdomen of each subject was exposed to a 4+ erythema dose of sunlight. Results were read 24 hours after the last dosage of medication and one month later.

Results

Although each subject developed a 4+ erythema reaction following the first dose of methoxsalen or placebo, the subsequent erythema reaction and pigmentation varied from subject to subject, over a period of 7 days during which time the medication was being administered. The intensity of the erythema reaction and pigmentation were such as to preclude any correlation between the experimental and the control group. The erythema and pigmentation reactions which appeared following the sunlight exposure given one month later showed different degrees of reactivity which could not be related to either placebo or drug ingestion.

TABLE III

Effect of sunlight on skin of patients taking single large dose 50 mg. of methoxsalen

	Intense Sunburn Reaction (4+)		Mild to Moderate Sunburn Reaction (1+ to 3+)	
	Placebo	50 mg. methoxsalen	Placebo	50 mg. methoxsalen
Number of subjects	3	4	12	11

EXPERIMENT #9

Purpose

To determine the effect of sunlight on the skin of patients taking a single large dose (50 mg.) of methoxsalen.

Method

Fifteen (15) men received a single 50 mg. dose of methoxsalen, and 15 received placebo medication. Two and one-half (2½) hours after drug ingestion, the abdomen of each was exposed to ½ hour of strong sunlight.

Results

The administration of a single large dose of methoxsalen produced an erythema reaction which did not differ significantly from a comparably exposed placebo group, (table III).

EXPERIMENT #10

Purpose

To determine the effectiveness of methoxsalen in protecting the skin from ultraviolet light "burn", using an artificial light source.

Method

The minimal exposure of ultraviolet light from an FS40-12 lamp required to produce a 4+ erythema response was determined for each of 50 men.

Thirty (30) men received 30 mg. of methoxsalen, and 20 men received placebo medication.

TABLE IV

Clinical reactions in patients from whom biopsy specimens were taken. (Exp. 10)

Subject	Medication	Pigmentation Remaining in Previously Exposed Side 1 Month After Exposure	Degree of Erythema 24 Hours After 4+ Erythema Exposure to Previously Exposed Half of Abdomen	Degree of Erythema 24 Hours. After 4+ Erythema Exposure to Previously Unexposed Half of Abdomen	Comment
1	Placebo	0	0	4+	Previously exposed half *protected* from further burn.
2	Placebo	3+	0	4+	Previously exposed half *protected* from further burn.
3	Methoxsalen (30 mg.)	2+	0	4+	Previously exposed half *protected* from further burn.
4	Methoxsalen (30 mg.)	4+	0	4+	Previously exposed half *protected* from further burn.
5	Placebo	4+	0	4+	Previously exposed half *protected* from further burn.
6	Placebo	2+	4+	4+	Previously exposed side *not* protected.
7	Methoxsalen (30 mg.)	2+	4+	4+	Previously exposed side *not* protected.
8	Methoxsalen (30 mg.)	1+	4+	4+	Previously exposed side *not* protected.

Two hours later a 4+ erythema exposure was administered to half the abdomen of each subject. Thereafter each man received this medication with exposure to the same half of the abdomen daily for a total of 7 days.

Four weeks later, a 4+ erythema exposure was given to the entire abdomen. Two biopsy specimens (one from each side of the abdomen) were obtained from each of eight (8) subjects. These 8 were selected from the total group of 50 men since they illustrated the variations in the clinical reactions which were noted in the total group, (table IV).

Results

(1) Although *all* subjects developed a 4+ erythema on the previously unexposed side of the abdomen, the previously exposed side showed varying degrees of erythema from 0 to 4+ of such a distribution as to indicate that there was no correlation between the degree of protection and the ingestion of methoxsalen or placebo (table V).

(2) Patients who ingested psoralens and were exposed to ultraviolet light did not develop more thickening of the stratum corneum nor did they have more melanocytic activity or pigment retention in the supra-basilar layers of the epidermis than non-psoralen, ultraviolet light exposed patients. The details of this study will be presented in another paper.

TABLE V

Degree of protection from "burn" (Exp. 10)

(Complete protection = no visible erythema partial protection = 1+ or 2+ erythema; no protection = 3+ or 4+ erythema.)

Medication and Dose	Number of Subjects	Complete Protection	Partial Protection	No Protection
Methoxsalen 30 mg.	30	18	5	7
Placebo	20	14	2	4

DISCUSSION

There were a total of 328 subjects used in these experiments; 208 were experimental (active medication) cases and 120 were controls (placebo medication). These subjects were given doses of

medication and doses of ultraviolet radiation (both artificial and natural) covering various possible clinical contingencies. In not one group could it be demonstrated that methoxsalen possessed any *regular* effect in influencing the erythema or pigmentary reactions of the skin.

Based on the observations of many physicians it is possible that methoxsalen may produce pronounced erythema reactivity in an occasional patient, but if this is so, it must be quite unusual, because our investigations failed to turn up such a result in a significant number of cases. In all we do not feel that methoxsalen can be depended upon to influence erythema production, pigmentogenesis, or to produce increased tolerance to ultravoilet light exposure.

The effect of ultraviolet light on normal human skin is well known. Following exposure to the sunburn radiations (2900–3100 Å) there is a latent period which ends with the appearance of erythema and subsequent pigmentation. These reactions vary in degree in different individuals Even a small exposure may suffice at times to protect some persons from further burn for *many months*, though no increased pigmentation is clinically apparent.

Not only are there variations in reactions to ultraviolet light in different individuals but even the same individual exhibits different responses on different parts of the skin or on the same part of the skin at different times.

These individual differences in response to ultraviolet light exposure make experimental results difficult to interpret. However, the use of sufficient numbers of experimental and control subjects provides a better basis for the evaluation of these reactions and we believe it also will resolve the apparently conflicting reports of various investigators.

Melanin formation is not an essential but an accessory factor in ultraviolet protection. We agree with Fitzpatrick and Szabo (5), that "on the basis of our present knowledge, it is not possible to assign relative values to the stratum corneum and malanin as biological ultraviolet filters". The mechanism involved in protection acquired by repeated exposures of ultraviolet light is not yet known.

SUMMARY AND CONCLUSIONS

(1) No alteration was observed in the amount of ultraviolet light, either from artificial or natural sources, required to produce minimal erythema in psoralen treated subjects as compared to placebo treated controls.

(2) Methoxsalen ingestion neither inhibited nor enhanced sunburn erythema or pigmentation.

(3) There were no consistent histologic differences in the skin of subjects taking methoxsalen, versus placebo, either in sunburned or in pigmented areas.

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